# The Chromosomes of Lycopodium lucidulum

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The Shining Fir-moss, *Lycopodium lucidulum* Michx. *(Huperzia lucidula* (Michx.) Trevisan), is an eastern North American endemic which forms colonies in moist coniferous and deciduous woods as well as swamps. The upright, isodichotomous stems root along their length as they become decumbent with age. The evergreen leaves, which are broadest above the middle, have stomates only on the undersides, and are usually coarsely serrate. They are produced in zones of longer and shorter leaves roughly corresponding to sterile and fertile leaves. Kidney-shaped sporangia are produced adaxially on the leaf base, and propagation organs (gemmae) develop at the end of each year's growth. In areas of overlap between *L. lucidulum* and the related northern species *L. selago* L. *(Huperzia selago* (L.) Bernh. ex Schrank & Mart.), intermediate sterile hybrids are occasionally found with either or both parents. These hybrids can be distinguished from *L. lucidulum* by their abortive spores and the presence of stomates on both surfaces of the leaves.

The chromosome number of *Lycopodium lucidulum* Michx. has been reported several times from North America and India. The counts from India (Mehra & Verma, 1957; Ninan, 1958) based on *Lycopodium lucidulum sensu* Clark (1880), however, are actually from a distinct species, *Lycopodium herterianum* Kümmerle (*Huperzia herteriana* (Kümm.) Sen & Sen, *L. sikkimense* Herter 1909 *non* K. Muell. 1861), and not the nearctic *Lycopodium lucidulum* Michx. Löve and Löve (1958) first reported $2n=264$ for *Lycopodium lucidulum* from Quebec, but without an accompanying photograph, a drawing, locality data, or a voucher specimen citation. Later (1965), they reported chromosome counts from Mt. Washington, New Hampshire (without citation of voucher) of *Lycopodium lucidulum* and *L. selago* (which they made into subspecies of *Huperzia selago*) remarking ". . . it has been at last possible to make exact counts of the chromosomes of material of ssp. *selago* with appressed and patent leaves from Mt. Washington and also ssp. *lucidula* from lower levels of that mountain. The chromosome number arrived at is $2n=272$."

The first indication that a lower ploidal level existed in *L. lucidulum* came from fertile material collected in Minnesota (Clearwater Co., Twin Lakes, 16 June 1977, *Wagner 77303*, MICH) and examined by F. S. Wagner. It appeared to have either 66, 67, or 68 pairs of chromosomes at meiosis, but the exact number of pairs could not be determined. A similar low count was obtained by J. M. Beitel in fertile material from Michigan (Livingston Co., Schwark Woods, W side of Merrill Road just S of Sheldon Road; T1N, R5E, SW sec. 35, 20 June 1979, *Beitel 79041*, MICH), which yielded approximately 66–69 pairs at meiosis. Material of *L. lucidulum* was collected in an acidic conifer swamp in Gray Co., Ontario (Osprey Township, 0.8 miles W of Rt. 24 on the S side of road dividing Concession 2 and 3) on 25 July 1981 by J. Beitel, W. H. Wagner and F. S. Wagner (*Beitel 81024*, MICH). The possibility of confusion with *L. selago* or its hybrids was ruled out by the presence of well formed spores and the lack of stomates on the adaxial leaf

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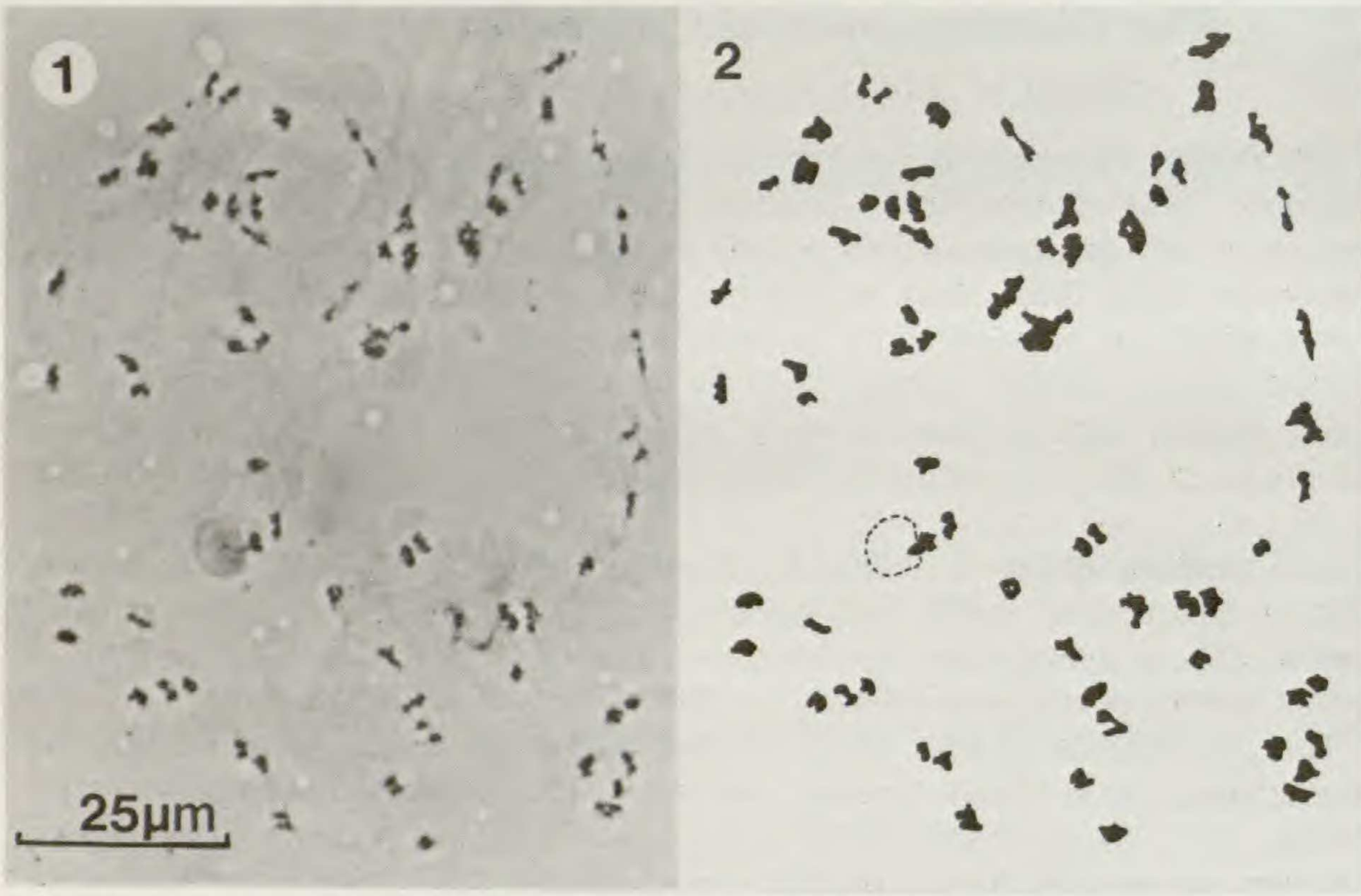


FIG. 1. Photograph of meiotic chromosomes of *Lycopodium lucidulum* Michx. (*Beitel 81024*), $n=67$.
FIG. 2. Interpretation of chromosome figure (dotted circle = nucleolus).

surfaces. The material was kept in a coldroom for two days, then submerged in distilled water in a clear plastic box kept at room temperature in the light for three days. Shoot tips bearing young, developing sporangia were placed in a saturated aqueous solution of para-dichlorobenzene and refrigerated at ca. 4°C for 24 hours. This use of a PDB treatment for condensing the meiotic chromosomes prior to fixation by Mehra and Verma (1957), Wilce (1965) and F. Wagner (1980) has reduced some of the problems involved in obtaining interpretable squashes. Excess water was blotted from the tips which were then placed in Newcomer's solution (Newcomer & Brant, 1954), left at room temperature for two hours, and placed in the freezer. Acetocarmine was used as a stain and squashing medium, Hoyer's solution was used for permanent mounting, and the slides were ringed with Eukitt plastic mounting medium.

Diakinesis was found to be the best stage for counting as the Metaphase I chromosomes, although darkly stained and well outlined, have the unfortunate tendency to clump. During diakinesis it was possible to discern the homologous chromosomes of each pair. Those undergoing early disjunction were coupled by a thin chromatin strand (as reported by Wilce, 1965). *Figures 1* and *2* clearly show a complement of $n=67$. Two large chromosome pairs were visible in all the figures examined. They can be seen slightly above the center of the photograph; the lower pair has a characteristic satellite. The nucleolus is seen slightly below and to the left of center. In over fifty figures observed and drawn, none exhibited a higher ploidal level than this figure.

The count of $n=67$ represents a new chromosome number for *Lycopodium lucidulum* and a new base number for the *L. selago* group of the segregate genus *Huperzia*. It is almost one-half the numbers previously reported from North America ($2n=264$, 272), but without voucher specimens or photographs of the chromosomes those records cannot be confirmed as to taxon or interpretation of the figures obtained. The previous low number of $n=$ ca. 68 reported by Löve and Löve (1961) for *Huperzia selago* ssp. *appressa* was discounted by them (1965) as ". . . most likely counted on an admixture of roots of a species of *Lycopodium* s. str. . . ."

The base number of $n=67$ fits into the gametophytic denominator scheme of Wagner and Wagner (1980) for *Lycopodium s. l.* based on multiples of 11 plus one aneuploid addition. Thus, $n=78$ of *Lycopodiella* equals $(7\times11)+1$, $n=34$ of *Lycopodium s. s.* equals $(3\times11)+1$, and $n=23$ of *Diphasiastrum* equals $(2\times11)+1$. This, however, does not explain the numbers reported for *L. cernuum* and the various epiphytic species (Löve, Löve & Pichi-Sermolli, 1977), nor for *L. carolinianum* (Bruce, 1976). The field of chromosome numbers in *Lycopodium s. l.* remains an open one with many species yet to be counted and many existing counts in need of confirmation.

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